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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
10/092,227	03/05/2002	Richard R. Bott	GC723	8688

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JANET KAISER CASTANEDA
GENENCOR INTERNATIONAL, INC.
925 PAGE MILL ROAD
PALO ALTO, CA 94304-1013

EXAMINER	
KERR, KATHLEEN M	

ART UNIT	PAPER NUMBER
1652	

DATE MAILED: 05/14/2004

Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No.	Applicant(s)	
	10/092,227	BOTT ET AL.	
	Examiner	Art Unit	
	Kathleen M Kerr	1652	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *01 March 2004*.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-18* is/are pending in the application.

4a) Of the above claim(s) *1-13* is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *14-18* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☒ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☒ Information Disclosure Statement(s) (PTO-1449 or PTO/SB/08) Paper No(s)/Mail Date *8/29/03*.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other: _____.

DETAILED ACTION

Application Status

1. In response to the previous Office action, a written restriction requirement (mailed on January 29, 2004), Applicants filed a response received on March 1, 2004. Claims 1-18 are pending in the instant Office action.

Election

2. Applicant's election of species "a", drawn to methods using crystal structures, in a paper received on January 29, 2004 is acknowledged. Because applicant did not distinctly and specifically point out the supposed errors in the restriction requirement, the election has been treated as an election without traverse (M.P.E.P. § 818.03(a)).

Claims 14-18, the elected species as noted in the previous Office action, will be examined herein.

Priority

3. No claims to priority for previous applications have been requested in the instant application. Therefore, this Office action considers prior art before the filling date of March 5, 2002.

Information Disclosure Statement

4. The information disclosure statement filed on August 29, 2003 has been reviewed, and its references have been considered as shown by the Examiner's initials next to each citation on the attached copy.

Compliance with the Sequence Rules

5. According to a statement filed on July 30, 2002, a paper copy of the sequence listing was filed with the instant application; however, none can be found in the filed. A new paper copy of the sequence listing is required as well as a statement that it is the same as the computer readable form filed on July 30, 2002 and that no new matter has been added.

Objections to the Specification

6. The specification is objected to because the title is not descriptive of the elected species to which the claims have been limited to herein. A new title is required that is clearly indicative of the invention to which the elected claims are drawn (see M.P.E.P. § 606.01). The Examiner suggests the following new title:

---High Throughput Mutagenesis Screening Method Using Enzyme Crystal Structures and Iterative Site Saturation Mutagenesis---

7. In the specification, the Abstract is objected to for not completely describing the disclosed subject matter (see M.P.E.P. § 608.01(b)). It is noted that in many databases and in foreign countries, the Abstract is crucial in defining the disclosed subject matter, thus, its completeness is essential. The Examiner suggests the inclusion of the use of the *Pseudomonas mendocina* cutinase.

Claim Objections

8. Claims 14 and 18 are objected to for having arduous claim structure. The Examiner suggests using a list, such as a), b), c), etc. to define the 6 method steps of the claim for clarity.

Claim Rejections - 35 U.S.C. § 112

The following is a quotation of the second paragraph of 35 U.S.C. § 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

9. Claims 14-18 are rejected under 35 U.S.C. § 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. In Claims 14 and 18, the iteration step is unclear. The concept of "repeat" site-saturation is confusing. It is the Examiner's understanding that saturation mutagenesis produces all possible mutations so it is unclear how repeating the process is at all useful. Perhaps the dual meaning of the word "site" in both "site-saturation mutagenesis", which in the art means changing a particular position to each of the other 19 amino acid residues, and "site" in other portions of the claim, which can mean general locations, like active site, binding site, etc. which can encompass several amino acid residues. Moreover, the template produced from feedback using the grading system does not necessarily indicate new sites for mutagenesis; is the 3D rendition used again in this step? Clarification is required.

10. Claims 14-18 are rejected under 35 U.S.C. § 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. The terms "three-dimensional rendition" and "three-dimensional model" are confusing as they are used in two different claims, Claims 14 and 18. Is there any difference in definition of the terms? In the art, three-dimensional crystal (or x-ray) structures are known for *F. solani* cutinase; said structures are high resolution (<2.0 angstroms). Is either a "rendition" or a "model" to mean this high-resolution type structure? Rendition implies more "low resolution" than model, but any such limitation is unclear. Clarification is required.

Claim Rejections - 35 U.S.C. § 102/103

The following is a quotation of the appropriate paragraphs of 35 U.S.C. § 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless –
>
> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

The following is a quotation of 35 U.S.C. § 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

11. Claims 14-15 and 17 are rejected under 35 U.S.C. § 102(b) as anticipated by or, in the alternative, under 35 U.S.C. § 103(a) as obvious over Poulouse (USPN 5,352,594) in view of Short (USPN 6,171,820, see IDS). The instant claims are drawn to methods for obtaining a variant cutinase by a method comprising (a) identifying residues for site-saturation mutagenesis using a 3D rendition of the cutinase, (b) making all 19 mutants at the position of said residues, (c) screening said mutants for substrate activity and grading said mutants wherein desirable mutants are used as templates for step d, and (d) repeating said mutagenesis at new sites using the templates.

Poulouse teaches a method for making mutant enzymes with altered substrate activities (see Abstract). A preferred enzyme is cutinase from *P. mendocina* whose sequence (see columns 4-5, bridging) is a truncated form of Applicant's SEQ ID NO:1. Selection of mutation site(s) is performed by choosing residues within 15 angstroms of the active site or at positions within 6

amino acids of active site residues (see column 5). In the examples of Poulouse, identification of the active site (the catalytic triad) of the cutinase was performed using sequence homology to known serine hydrolases with known 3D structures (see columns 9-10). Poulouse teaches producing mutants and assaying for activity (see column 6, lines 23-39 and columns 11-20, Tables). Poulouse also teaches saturating selected sites on the enzyme to achieve the best results (see column 6, lines 40-47) and teaches "substitutions at more than one site within the parameters of the invention ... in order to **further optimize** the results" (emphasis added) (see column 6, lines 47-49). Thus, Poulouse teaches an iterative process as required by the instant claims.

In the occasion that the phrase "further optimize" is not considered to describe an iterative process, such iteration would have been obvious in view of Short. Short teaches methods of directing evolution of enzymes using saturation mutagenesis, preferably in an iterative manner combined with screening (see column 35, lines 15-20 and columns 55-56, Example 5).

At the time of the invention, methods of producing variant enzymes using (a) site-saturation mutagenesis, as indicated by structural data, (b) screening, and (c) repeating with screened products having desirable activities was obvious because defining a particular single-site mutant as useful and using said single-site mutant for further mutagenesis producing even more useful double mutants is an efficient use of time and resources as opposed to making all possible combinations of double mutants first. One would have been motivated to practice said iteration using time- and resource-saving practices because said practices are practical laboratory techniques. One would have had a reasonable expectation of success in production of double,

etc. mutants by an iterative process because the process is identical except for the template. Moreover, the Tables in columns 11-18 of Poulouse evidence producing single mutants and using the most desirable single mutants for production of double mutants.

12. Claims 16 and 18 are rejected under 35 U.S.C. 103(a) as being unpatentable over Poulouse (USPN 5,352,594) in view of Abo *et al.* (WO 00/34450) and optionally in view of Short (USPN 6,171,820, see IDS). The instant claims are drawn to methods for obtaining a variant *Pseudomonas* cutinase by a method comprising (a) identifying residues for site-saturation mutagenesis using a 3D model of the cutinase, (b) making all 19 mutants at the position of said residues, (c) screening said mutants for polyesterase activity and thermostability and grading said mutants wherein desirable mutants are used as templates for step d, and (d) repeating said mutagenesis at new sites using the templates.

Poulouse and Short teach as described above. Poulouse also teaches assaying for polyesterase activity (see columns 11 and 12). Poulouse does not teach screening for thermostability of the variant cutinases.

Abo *et al.* teach producing thermostable variant cutinase that retain their catalytic activity with a polyester substrate, BETEB (see pages 17-21).

At the time of the invention, it would have been obvious to practice the methods taught by Poulouse, and optionally Short, to produce better cutinases wherein better is equivalent to more active against a polyester substrate and more thermostable because said mutants are expressly proposed by Abo *et al.* although produced by different means. One would have been motivated to combine the teachings of Poulouse, optionally Short, and Abo *et al.* because of the

commercial value of thermostable, active cutinases as noted by Abo *et al.* (see page 1). One would have had a reasonable expectation of success that screening cutinases using the methods of Poulouse and optionally Short would alter both the catalytic activity with a particular substrate as well as the thermostability of the cutinases because both kinds of mutant variants have been found independently by Poulouse and Abo *et al.* respectively.

Conclusion

13. Claims 14-18 are not allowed for the reasons identified in the numbered sections of this Office action. Applicants must respond to the objections/rejections in each of the numbered sections in this Office action to be fully responsive in prosecution.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Kathleen M Kerr whose telephone number is (571) 272-0931. The examiner can normally be reached on Monday through Friday, from 9:00am to 6pm.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Ponnathupura Achutamurthy can be reached on (571) 272-0928. The fax phone number for the organization where this application or proceeding is assigned is 703-872-9306.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free).

Kathleen M Kerr
Examiner
Art Unit 1652

May 13, 2004